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EXAMINER
STEADMAN, DAVID J

ART UNIT	PAPER NUMBER
1652	20

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Please find below and/or attached an Office communication concerning this application or proceeding.

PTO-90C (Rev. 10/03)

FILE COPY

Office Action Summary	Application No.	Applicant(s)	
	09/815,533	ARINI ET AL.	
	Examiner	Art Unit	
	David J Steadman	1652	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *13 July 2003* .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *81-92* is/are pending in the application.

4a) Of the above claim(s) *91 and 92* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *81-90* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☒ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449) Paper No(s) _____ .
4) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: .

DETAILED ACTION

Status of the Application

[1] A request for continued examination under 37 CFR 1.114, including the fee set forth in 37 CFR 1.17(e), was filed in this application after final rejection. Since this application is eligible for continued examination under 37 CFR 1.114, and the fee set forth in 37 CFR 1.17(e) has been timely paid, the finality of the previous Office action has been withdrawn pursuant to 37 CFR 1.114. Applicant's submission filed on July 17, 2003, has been entered.

[2] Claims 81-92 are pending in the application.

[3] Applicant's amendment to the specification in Paper No. 19, filed July 17, 2003, is acknowledged.

[4] Receipt of a computer readable form (CRF) of the sequence listing, a paper copy thereof, and a statement that the paper copy and CRF of the sequence listing are identical in Paper No. 19 is acknowledged.

[5] Applicant's cancellation of claims 69-80 and addition of claims 81-92 in Paper No. 16, filed May 15, 2003, is acknowledged.

[6] Receipt of a Declaration under 37 CFR 1.132 filed May 15, 2003 as Paper No. 16 is acknowledged.

[7] Applicant's arguments filed in Paper Nos. 16 and 19 have been fully considered and are deemed to be persuasive to overcome some of the rejections and/or objections previously applied. Rejections and/or objections not reiterated from previous office actions are hereby withdrawn.

[8] The text of those sections of Title 35 U.S. Code not included in the instant action can be found in a prior Office action.

Election/Restriction

[9] In the amendment of Paper No. 16, claims 81-92 have been added. Claims 81-90, drawn to a process for the production of recombinant catalytically active tc-uPA, correspond to elected Group I of the restriction as set forth in Paper No. 8, mailed February 21, 2002. Claims 91-92 are drawn to a process for purifying recombinant high molecular weight (HMW) two-chain urokinase plasminogen activator (tc-uPA) corresponding to non-elected Groups II and III of the restriction as set forth in Paper No. 8. In the advisory action of Paper No. 17, the examiner indicated that these claims are drawn to a non-elected invention and would not be co-examined with the claims of the elected invention (see item 5 of Paper No. 17). At page 3 of the amendment filed July 17, 2003, applicant argues claims 91-92 are dependent upon claim 88 and since claims 91-92 contain the recitations of claim 88 of elected Group I, it is believed that no undue burden is necessary to co-examine claims 91-92 with the claims of the elected invention. Applicant requests that no election restriction be required. Applicant's argument is not found persuasive.

MPEP § 803 states that a serious burden on the examiner may be prima facie shown if the examiner shows by appropriate explanation either separate classification, separate status in the art, or a different field of search. In the instant case, the claims of elected Group I and claims 91-92 as presented have separate classification (see the

classifications of Groups I-III in the Office action of Paper No. 8). Furthermore, claims 91-92 recite specific limitations, e.g., purification using a cation exchanger using buffer solutions at pH 5.5 to 6.5 and 6 to 7.5, that are not recited in claims 81-90, and therefore, a different field of search would be required to search claims 91-92.

[10] The requirement is still deemed proper and is therefore made FINAL.

[11] Claims 91-92 are withdrawn from further consideration pursuant to 37 CFR 1.142(b), as being drawn to nonelected inventions, there being no allowable generic or linking claim.

Specification

[12] Applicant's amendment to the specification, i.e., "--This Application is a continuation of Serial No. 09/815,533, filed March 16, 2001.--", as set forth at page 2 of the amendment filed July 17, 2003, is improper. As a request for continued examination application maintains the same application number, the instant application cannot claim priority to itself. It is suggested that applicant delete this amendment.

Claim Objections

[13] In view of applicant's amendment to the claims, the objections as set forth in items 3-5 in the Office action mailed February 12, 2003, are withdrawn.

Claim Rejections - 35 USC § 112, Second Paragraph

[14] In view of applicant's amendment to the claims, the rejections as set forth in items 6a-c in the Office action mailed February 12, 2003, are withdrawn.

[15] Claim 87 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 87 is drawn to the process of claim 81 wherein said cell line is cultured for a time of 48 to 200 hours. It is noted that the Declaration of Paper No. 16 indicates that a culturing time of at least 96 hours is required to produce the requisite 95% catalytically active tc-uPA. For example, the Figure 2 of the Declaration shows that at 48 hours, two-chain uPA is present at 92%, which is below the recited level of at least 95%. Thus, based on the evidence provided in the Declaration, it is unclear as to how a culturing time less than 96 hours will allow for the production of the required 95% catalytically active tc-uPA. It is suggested that applicant clarify the meaning of the claim.

Claim Rejections - 35 USC § 112, First Paragraph

[16] Claims 81-90 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a process for producing at least 95% catalytically active tc-uPA as recited in claim 81, wherein the eukaryotic cell line is cultured for a time of at least 96 hours, does not reasonably provide enablement for a process for producing at least 95% catalytically active tc-uPA as recited in claim 81, wherein the eukaryotic cell line is cultured for a time of less than 96 hours. The specification does

not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention commensurate in scope with these claims.

It is the examiner's position that undue experimentation would be required for a skilled artisan to make and/or use the entire scope of the claimed invention. Factors to be considered in determining whether undue experimentation is required are summarized in *In re Wands* (858 F.2d 731, 737, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988)) as follows: (A) The breadth of the claims; (B) The nature of the invention; (C) The state of the prior art; (D) The level of one of ordinary skill; (E) The level of predictability in the art; (F) The amount of direction provided by the inventor; (G) The existence of working examples; and (H) The quantity of experimentation needed to make or use the invention based on the content of the disclosure. See MPEP § 2164.01(a). The Factors most relevant to the instant rejection are addressed in detail below.

- The claims are overly broad in scope: The claims are so broad as to encompass a process for producing at least 95% catalytically active tc-uPA as recited in claim 81, wherein the eukaryotic cell line is cultured for less than 96 hours. The scope of the claims is not commensurate with the enablement provided by the disclosure with regard to the time of culture of the cells. In this case, the disclosure, in light of the evidence provided by applicant in the Declaration filed May 15, 2003, is limited to a process for producing at least 95% catalytically active tc-uPA as recited in claim 81, wherein the eukaryotic cell line is cultured for at least 96 hours.

- The lack of guidance and working examples: The Declaration under 37 CFR 1.132 submitted as part of Paper No. 16 indicates that a culturing time of at least 96 hours is required to obtain 95% catalytically active tc-uPA (see page 2, Figure 2 of the Declaration of Paper No. 16). Therefore, one of skill in the art would recognize that at least 96 hours of cell culturing is required to obtain the recited level of catalytically active tc-uPA and the specification fails to teach how to obtain such a level using culturing times of less than 96 hours.

- The amount of experimentation required is undue: Based on the Declaration of Paper No. 16, one of skill in the art would have no expectation of obtaining 95% catalytically active tc-uPA using culturing times of less than 96 hours. Thus, one of skill in the art would recognize that an undue amount of experimentation would be required to achieve at least 95% tc-uPA by culturing cells less than 96 hours.

Thus, applicant has not provided sufficient guidance to enable one of ordinary skill in the art to make and use the claimed invention in a manner reasonably correlated with the scope of the claims. The scope of the claims must bear a reasonable correlation with the scope of enablement (*In re Fisher*, 166 USPQ 19 24 (CCPA 1970)). Without sufficient guidance, determination of having the desired biological characteristics is unpredictable and the experimentation left to those skilled in the art is unnecessarily, and improperly, extensive and undue. See *In re Wands* 858 F.2d 731, 8 USPQ2nd 1400 (Fed. Cir, 1988).

Beginning at the bottom of page 8 of the amendment filed May 15, 2003, applicant argues the rejection has been overcome by amendment. Applicant's argument is not found persuasive.

In view of the evidence submitted by applicant in the Declaration filed May 15, 2003, the scope of enablement rejection has been re-written to address the enablement provided by the specification in light of the Declaration. Accordingly, the claims do not enable the entire scope of claimed methods as stated above.

Claim Rejections - 35 USC § 102

[17] In view of applicant's cancellation of claims 69 and 70, the rejection of claims 69 and 70 under 35 U.S.C. 102(b) as being anticipated by Okabayashi et al. (*Cell Struct Funct* 14:579-586) is withdrawn in view of applicant's amendment to claim 81 to limit the tc-uPA produced to at least 95% catalytically active tc-uPA and the Declaration under 37 CFR 1.132 showing that under the conditions of Okabayashi et al. only 67% of the urokinase is catalytically active tc-uPA (see page 10 of Paper No. 16 and page 2 of the Declaration of Paper No. 16). Therefore, the reference of Okabayashi et al. does not anticipate the claimed invention.

Claim Rejections - 35 USC § 103

[18] Upon reconsideration of the rejections under 35 USC 103(a) as set forth in items 10-11 of the Office action mailed February 12, 2003, the rejections have been withdrawn. It is noted that the rejections have been NOT been withdrawn in view of

applicant's arguments and instead have been withdrawn in favor of the rejections as stated in items 18 and 19 below. As the rejections as stated in items 18 and 19 below no longer include the references of Nobuhara et al., Miwa et al., and Hu, applicant's arguments, to the extent said arguments address rejections over these references, are rendered moot. Applicant's arguments, to the extent said arguments address the reference of Okabayashi et al., have been addressed below.

[19] Claims 81-85 and 87-89 are rejected under 35 U.S.C. 103(a) as being unpatentable over Okabayashi et al. (*Cell Struc Funct* 14:579-586) in view of the state of the art as represented by Zang et al. (*Biotechnology (NY)* 13:389-392). Claim 81 is drawn to a process for the production of recombinant catalytically active two-chain urokinase (tc-uPA) by culturing a eukaryotic cell line transfected with a cDNA encoding a urokinase precursor in the presence of an alkanoic acid (as encompassed by claim 81) resulting in the production of at least 95% catalytically active tc-uPA. Claim 82 limits the cell line of claim 81 to CHO or CHO-Messi cells. Claim 83 limits the culture medium of claim 81 to a serum-free medium. Claim 84 limits the concentration of alkanoic acid of claim 81 to a range of 0.1 to 20 mM. Claim 85 limits the temperature of culturing after addition of alkanoic acid to 30 to 37 degrees Celsius. Claim 87 limits the time of culturing to a range of 48 to 200 hours. Claim 88 is drawn to the method of claim 81 further comprising a step of recovering the cell culture medium. Claim 89 limits the production level of tc-uPA to at least 4000 IU/ml culture medium.

Okabayashi et al. disclose a method for the expression of recombinant human urokinase using Chinese hamster ovary (CHO) cells cultured in a medium comprising

sodium n-butyrate (page 579, abstract). Okabayashi et al. teach the CHO cells were transfected with an expression vector comprising a cDNA encoding human pre-pro-urokinase (page 580). Okabayashi et al. teach analysis of a sample of the culture medium following a 24-hour period for urokinase activity (page 582, Table 1 caption). This analysis revealed that addition of butyrate in the CHO cell culture medium results in a significant increase in urokinase activity relative to CHO cells cultured without butyrate (page 582, Table 1). Because pre-pro-urokinase and pro-urokinase are known in the art to be catalytically inactive, the urokinase activity present in the collected culture medium is two chain urokinase (tc-uPA). Okabayashi et al. do not teach a culturing time of 48 to 200 hours or that the urokinase produced by their method is at least 95% tc-uPA.

At the time of the invention, recombinant production of urokinase using CHO cells as an expression host was well known in the art. This is exemplified by the reference of Zang et al. who generally teach a method for the production of recombinant urokinase in CHO cells using a serum-free cell culture medium. In particular, Zhang et al. teach a culturing time of up to five days is sufficient for a near-maximum concentration of urokinase (see page 390, Figure 3A). Zang et al. teach at least one advantage of using serum-free medium in the recombinant production of urokinase is a reduction in the likelihood of introducing transmissible contaminants (page 389, left column).

Therefore, at the time of the invention, it would have been obvious to one of ordinary skill in the art to combine the teachings of Okabayashi et al. and Zang et al. for

a method of recombinantly producing urokinase by culturing CHO cells in a serum-free medium as taught by Zang et al. in the presence of sodium butyrate as taught by Okabayashi et al. for at least 5 days as taught by Zang et al. One would have been motivated for a method of recombinantly producing urokinase by culturing CHO cells in a serum-free medium in order to reduce the possibility of contamination as described by Zang et al., to culture the cells in the presence of sodium butyrate in order to significantly increase the production of urokinase as taught by Okabayashi et al., and to culture the CHO cells for at least 5 days in order to reach the near-maximum concentration of urokinase in the medium as taught by Zang et al. One would have a reasonable expectation of success for a method of recombinantly producing urokinase by culturing CHO cells in a serum-free medium in the presence of sodium butyrate for at least 5 days because of the results of Okabayashi et al. and Zang et al. Therefore, claims 81-85 and 87-89, drawn to a process for the recombinant production of tc-uPA as described above would have been obvious to one of ordinary skill in the art.

It is noted that the combined references do not expressly teach the production of tc-uPA at a level of at least 95%. However, based on the evidence provided in the Declaration filed May 15, 2003, culturing the CHO cells in the presence of butyrate for at least 4 days (96 hours) would inherently achieve the desired 95% level of catalytically active tc-uPA. The Declaration filed May 15, 2003 shows that, using the method of Okabayashi et al., only 67% tc-uPA is produced after 24 hours culturing in the presence of butyrate (see page 2, Figure 2). However, a longer culture time of at least 96 hours under these same conditions results in at least 95% tc-uPA. Therefore, one only need

follow the method as taught by the combined references of Okabayashi et al. and Zang et al. to culture the CHO cells in the presence of butyrate for 5 days in order to obtain at least 95% tc-uPA.

[20] Claims 86 and 90 are rejected under 35 U.S.C. 103(a) as being unpatentable over Okabayashi et al. in view of the state of the art as represented by Zang et al. as applied to claims 81-85 and 87-89 above, and further in view of Anderson et al. (US Patent 6,506,598). Claim 86 limits the culture temperature of the method of claim 81 to a range of 33 to 35 degrees Celsius. Claim 90 is drawn to the process of claim 88 wherein said culture medium is acidified with an acid of pH from 5 to 5.8 and optionally a non-ionic detergent is added and the culture medium filtered.

Okabayashi et al. and Zang et al. disclose the teachings as described above.

Anderson et al. teach a process for culturing CHO cells for the recombinant production of urokinase in a serum-free medium at a temperature of between 30 and 35 degrees Celsius in the presence of a butyrate salt in order to improve the occupancy of an N-linked glycosylation site in the produced urokinase (column 8, lines 16-22). Anderson et al. teach methods of recovering the produced urokinase in the culture medium (columns 18-20). For example, Anderson et al. teach centrifugation to remove the cells, loading of the culture medium onto a lysine affinity column, and elution of urokinase with a buffer of pH 5.0 (column 19).

Therefore, at the time of the invention, it would have been obvious to one of ordinary skill in the art to combine the teachings of Okabayashi et al., Zang et al., and Anderson et al. for a method of recombinantly producing urokinase by culturing CHO

cells in a serum-free medium in the presence of sodium butyrate for at least 5 days at a temperature of 30 to 35 degrees Celsius and recovering the urokinase from an affinity column by elution with a buffer of pH 5.0. One would have been motivated to culture the cells at temperature of 30 to 35 degrees Celsius in order to improve the occupancy of an N-linked glycosylation site as taught by Anderson et al. and to add a buffer of pH 5.0 to an affinity column comprising the urokinase in order to elute the bound urokinase. One would have a reasonable expectation of success for a method of recombinantly producing urokinase by culturing CHO cells in a serum-free medium in the presence of sodium butyrate for at least 5 days at a temperature of 30 to 35 degrees Celsius and recovering the urokinase from an affinity column with a buffer of pH 5.0 because of the results of Okabayashi et al., Zang et al., and Anderson et al. Therefore, claims 86 and 89, drawn to a process for the recombinant production of tc-uPA as described above would have been obvious to one of ordinary skill in the art.

Applicant argues (beginning at the bottom of page 9 of the amendment filed May 15, 2003) claim 81 has been amended to recite a process for the production of tc-uPA resulting in a production of at least 95% tc-uPA and this process is novel with respect to Okabayashi et al., which is a process for increasing overall urokinase production. Applicant argues claim 81 is also novel with respect to the urokinase produced by Okabayashi et al. as the declaration filed May 15, 2003 demonstrates that the method of Okabayashi et al. yields only 67% tc-uPA after a 24 hour cultivation time. Applicant argues the instant claims require at least 95% catalytically active tc-uPA, an amount

that is reached after a butyrate incubation time of 72-96 hours. Applicant states it is their opinion that the amendment of claim 81 to a preferred time of 48 to 200 hours incubation of the cells in the presence of butyrate could have contributed substantially to the novelty of former claim 71 with respect to the teachings of Okabayashi et al. Applicant argues (beginning at the bottom of page 11 of the amendment filed May 15, 2003) that the presence of butyrate triggers an unexpected activation of MMPs probably representing the conversion to at least 95% tc-uPA. Applicant argues this effect could not be triggered by the prior art. Applicant's argument is not found persuasive.

Based on an analysis of the teachings of Okabayashi et al. (as described above), the instant specification (pages 15-18), and the declaration filed May 15, 2003 (see particularly page 2, Figure 2), it appears that one merely need to continue cultivating the cells in the presence of butyrate using the method disclosed by Okabayashi et al. for a time of at least 96 hours to achieve the desired 95% catalytically active tc-uPA. This conclusion is supported by the declaration filed May 15, 2003 (see page 2, Figure 1), which shows that, using the method of Okabayashi et al., only 67% tc-uPA is produced after 24 hours culturing in the presence of butyrate. However, a longer culture time of at least 96 hours under these same conditions results in at least 95% tc-uPA. Therefore, one only need follow the method as disclosed by Okabayashi et al. and culture the cells in the presence of butyrate for at least 96 hours to obtain at least 95% tc-uPA. It is noted that Okabayashi et al. do not provide any reasoning as to why one of ordinary skill in the art would cease culture of the cells at 24 hours - as evidenced by the prior art, culture times of at least 5 days are known in the production of urokinase.

Conclusion

[21] Status of the claims:

- Claims 81-92 are pending.
- Claims 91-92 are withdrawn from further consideration.
- Claims 81-90 are rejected.
- No claim is in condition for allowance.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to David Steadman, whose telephone number is (703) 308-3934. The Examiner can normally be reached Monday-Friday from 7:00 am to 5:00 pm. If attempts to reach the Examiner by telephone are unsuccessful, the Examiner's supervisor, Ponnathapura Achutamurthy, can be reached at (703) 308-3804. The FAX number for submission of official papers to Group 1600 is (703) 308-4242. Draft or informal FAX communications should be directed to (703) 746-5078. Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the Art Unit receptionist whose telephone number is (703) 308-0196.

David J. Steadman
Patent Examiner
Art Unit 1652

10/03/03

DAVID STEADMAN
PATENT EXAMINER